A REVIEW OF NEMATOPHAGY BY TERRESTRIAL TARDIGRADES WITH NEW OBSERVATIONS ON SOME AUSTRALIAN SPECIES

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MACKNESS, B.S., 2003:06:15. A review of nematophagy by terrestrial tardigrades with new observations on some Australian species. *Proceedings of the Royal Society of Victoria* 115(1): 1-5. ISSN 0035-9211.

A literature review of nematophagy by terrestrial tardigrades was undertaken including a survey of 93 species of the genus *Macrobiotus* to determine whether there were any morphological criteria common to all nematophagous species. Preliminary results suggest that all nematophagous species have buccal tube widths greater than 17% of the tube length and that they also possess peribuccal lamellae. Two new nematophagous tardigrades, *Macrobiotus hieronimi* Pilato & Claxton, 1988 and *M. peteri* Pilato et al., 1989 have been recognized as well as further records of nematode predation by tardigrades from Australia.

Key words: Tardigrades, nematophagy, buccal tubes, peribuccal lamellae

TERRESTRIAL tardigrades are primarily vegetarian, although some are known to take animal food including nematodes (Ramazzotti & Maucci 1983). There are few reports of nematophagy however found in the literature. Linford & Oliveira (1938) reported three predaeious tardigrades (*Macrobiotus* spp.) attaeking root-knot nematodes *Heterodera* sp. (now various species of *Meliodogyne*). Le Gros (1958) stated that tardigrades attaeked nematodes in addition to rotifers and other tardigrades. Hutchinson & Streu (1960) reported an unidentified tardigrade feeding on the nematodes *Trichodorus* sp. and *Tylenchus* sp. in the United States and in Sri Lanka. Doneaster & Hooper (1961) and Doneaster (1962) recorded *Macrobiotus richtersi* Murray, 1911 piereing nematodes and suggested that a toxin might have been secreted to immobilize large nematodes sueh as mononehids. Esser (1963) and Esser & Sobers (1964) examined the possibility of tardigrades being used as biological control agents of nematodes. Maerobiotid tardigrades, in citrus orehards, have been recorded feeding on the nematodes *Tylenchulus* sp. and *Meloidogyne* sp. (Boosalis & Manakau 1965, Stirling & Mankau 1977).

Sayre (1969) cultured the tardigrade *Hypsibius* (=*Isohypsibius*) *myrops* (Du Bois-Reymond Mareus, 1944) on nematodes for several months. He also observed individuals feeding on the plant nematodes *Meloidogyne* sp. and *Ditylenchus* sp. Hallas & Yeates (1972) recorded *Macrobiotus harmsworthi* Murray, 1907 feeding on nematodes and reported a strong correlation between total number of nematodes and monthly tardigrade populations. Hallas & Yates (1972) also suggested that *Hypsibius* (*I.*) *myrops* (*sensu* Sayre 1969) might be identieal to *Hypsibius* (*I.*) *prosostomus*, which they also reported feeding on nematodes.

Several other authors have reported general ineidents of predation by tardigrades (Cayrol 1976, Morgan 1977, Small 1988). Further investigations of predation by tardigrades were undertaken by Sayre & Wergin (1979) using a seanning eleetron mieroseope. Esser (1990) summarized more than 25 years of observing nematodes and tardigrades. These observations primarily eoneerned hypsibiid tardigrades feeding on a variety of nematodes. Bird (1996) figured *Macrobiotus* ef. *pseudohufelandi* Iharos, 1966 eating an unidentified soil nematode.

Hallas & Yeates (1972: 289) eharaeterized predaeious tardigrades in having '... a mouth opening (oral aperture) whieh points direetly forward, a short, wide pharyngeal tube, strongly built stylets and a large bulbus'. A survey was undertaken of published buecal tube widths of 93 species of *Macrobiotus* to aseertain whether this eriterion eould be used as a guide to reeognizing pred-aeious tardigrades. The ineidence of buccal lamellae was also examined. The results of this survey are reported in Table 1. The author reports two hitherto unknown speeies of nematophagous tardigrades as well as further reeords of nematode predation by tardigrades from Australia.

Species	BTR	Length	Lamellae
M. beotiae Durante & Maucci, 1979	(25% of length)	880 µm	Yes
M. gildae Maucci & Durante Pasa, 1980	(22% of length)	500 µm	Yes
M. grandis Richters, 1911	(17% of length)	900 µm	Yes
***M. harmsworthi* Murray, 1907**	**(20% of length)**	**650 µm**	**Yes**
***M. hieronimi* Pilato & Claxton, 1988**	**(17% of length)**	**488 µm**	**Yes**
M. maucci Pilato, 1974	(22% of length)	450 µm	Yes
***M. peteri* Pilato et al. 1989**	**(17% of length)**	**690 µm**	**Yes**
M. pseplus du Bois-Reymond Marcus, 1944	(17% of length)	800 µm	Yes
***M. pseudohufelandi* Iharos, 1966**	**(18% of length)**	**510 µm**	**Yes**
M. recens Cuenot, 1932	(21% of length)	624 µm	Yes
***M. richtersi* Murray, 1911**	**(20% of length)**	**750-1000 µm**	**Yes**
M. snaresensis Horning *et al.*,1978	(22% of length)	450 µm	Yes
M. spectabilis Thulin, 1928	(17% of length)	700 µm	Yes

Table 1. Species of *Macrobiotus* with buccal tube widths greater than 17% of buccal tube length. (Source Ramazzotti & Maucci 1983 and references cited in text). Known nematophagous tardigrades highlighted in bold. (BTR = Buccal tube ratio)

Feeding by tardigrades

Tardigrades have a complex series of organs associated with feeding. These include the buccal orifice, stylets, the pharynx and the oesophagus. The oral aperture can be terminal as in most macrobiotids or slightly ventral as in most hypsibids. The buccal cavity opens with a circular ring that may support a crown of buccal lamellae (Fig 1A). These allow the mouth to be applied like a sucker to prey (Ramazzotti & Maucci 1983). There are piercing organs on either side of the mouth called stylets. Their composition remains unclear although Marcus (1927) and Kaestner (1968) suggest they are calcareous. Bird (1996) has recorded marked bi-refringence of the stylets under polarized light and suggested this may be due to their inferred crystalline nature. The stylets themselves, are operated by several retracting and protracting muscles. Tardigrades use these stylets to pierce the cuticles of their prey. The buccal cavity is followed by a buccal tube that extends into the pharynx. The pharynx functions as a sucking organ and is generally ovoid in shape (Ramazzotti & Maucci 1983). Food is not masticated but passed by sucking action through a short oesophagus into the midgut where digestion takes place.

In some tardigrades, particularly Family Macrobiotidae, there is a series of circular swellings called placoids in the pharyngeal bulb (Fig 1B). They are thought to play a role in mechanical reinforcement of the pharynx (Ramazzotti & Maucci 1983). There are also large rostral or salivary glands located lateral to the buccal tube and pharyngeal bulb. It has not been confirmed whether these glands secrete digestive juices and indeed some authors, such as Cuénot (1932), have ascribed an exeretory function to them.

There appears to be no pattern to the choice or size of nematode taken as prey by tardigrades (Doncaster & Hooper 1961). Tardigrades have been recorded passing within 10 µm of a nematode without any attempt to attack (Esser 1990). Likewise, not all attacks by tardigrades are successful. Esser (1990) observed an attack by a tardigrade on the nematode *Enchodelus* (three and a half times larger than the tardigrade) that was unsuccessful as well as another on a large dorylaim. Prey is normally pierced with the stylets and the body contents sucked out. Several tardigrades may attack the same nematode simultaneously (Doncaster & Hooper 1961).

Sometimes the prey is swallowed whole. Ramazzotti & Maucci (1983) observed *Macrobiotus richtersi,* with a buccal tube of 12 µm, with a whole nematode inside its stomach. Hallas & Yeates (1972) observed *M. harmsworthi* with a nematode tail projecting out of its cloaca. Apparently it had ingested the nematode tail first and the thinner posterior end had passed through the gut unruptured while the anterior end had been digested.

When tardigrade attacks occur, they can be quite rapacious and sustained. Esser & Sobers (1964: 333) reported an earlier observation by Esser of tardigrades

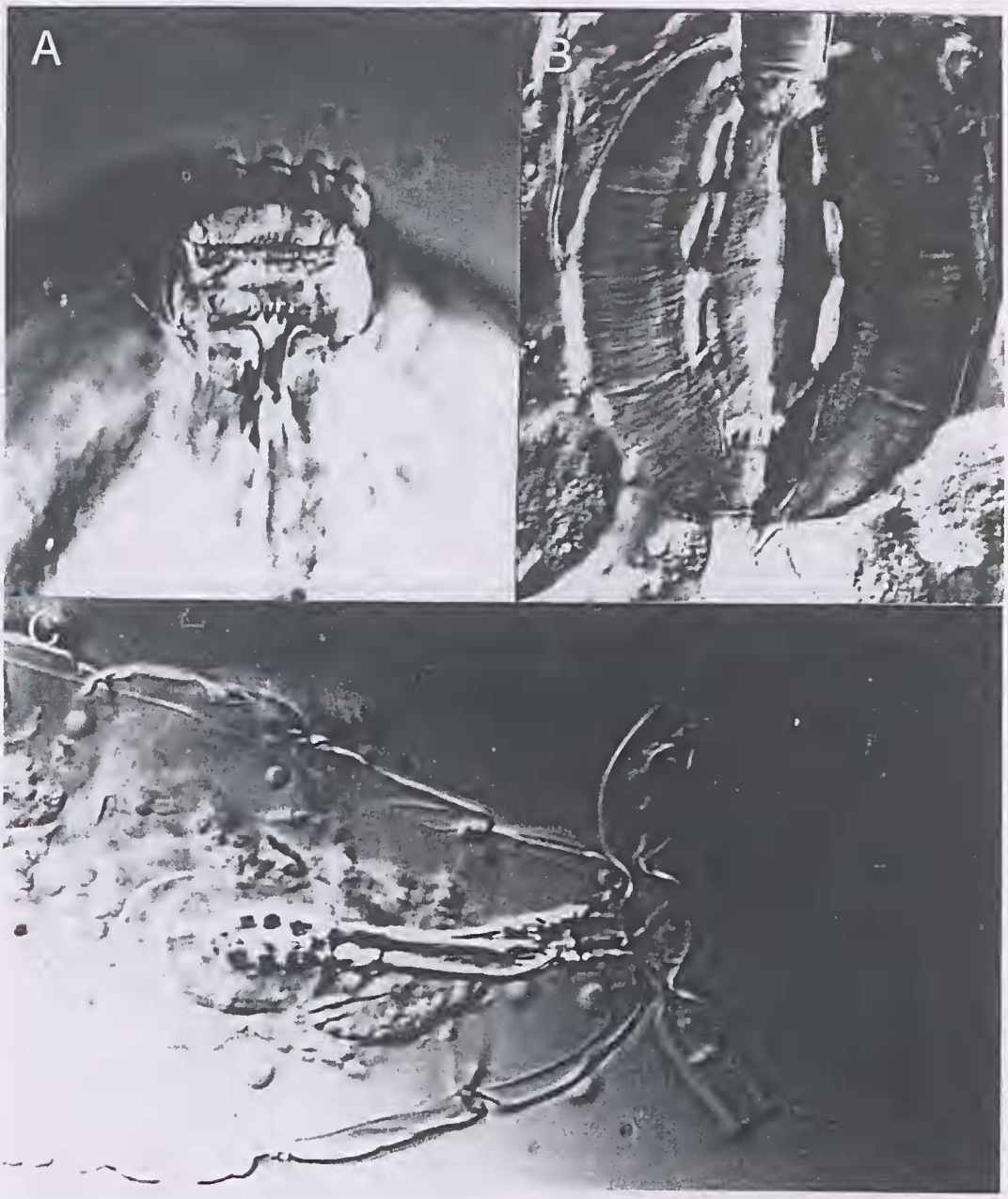

Fig. 1. A, Peribuccal lamellae at entrance to buccal cavity. B, Pharyngeal bulb with placoids. C, *Macrobiotus* sp. from Mt Ainsley, ACT with nematode. Scale bar = 100 μm

'viciously attaeking nematodes, ripping many of them to pieces and swallowing some of them whole'. Hutchinson & Streu (1960) reported that tardigrades made distinet tears in the eutieles of nematodes while Esser (1990) recorded a tardigrade holding on to a nematode for over three hours. Doncaster & Hooper (1961) suggested that tardigrades might inject a toxic secretion into nematodes in order to subdue them.

The following tardigrade speeies from Australia and Lord Howe Island have been observed feeding on nematodes.

Macrobiotus hieronimi Pilato & Claxton, 1988

One specimen (LH3-13) collected by Jo Mon from Lord Howe Island (6/7/80) and mounted by S.K. Claxton. Tardigrade is 420 µm long and recovered from damp moss *Ptychomitrium* sp. with 25-50% moisture content. The nematode is being attacked head on and all of the pharyngeal region has been consumed and internal structures dissolved. Consequently identification of the nematode is not possible. This is the first published record of nematophagy by this species.

Macrobiotus peteri Pilato et al., 1989

Three specimens (ATS1065-3, 1067-16, 1090-10) collected and mounted by S.K. Claxton, Douglas Park, NSW. Tardigrades found in moss *Dicnemoloma* sp. The nematodes cannot be identified. Tardigrades are 320 µm, 420 µm and 430 µm long respectively. This is the first published record of nematophagy by this species.

Macrobiotus richtersi Murray, 1911

One specimen (ATS 417-1) collected and mounted by S.K. Claxton, Cedarvale, NSW from moss/lichen. The nematode cannot be identified. The tardigrade is 420 µm long.

Milnesium tardigradum Doyère, 1840

One specimen (ATS 1058-37) collected by S.K. Claxton, Cambewarra Mountains, NSW. Tardigrade found in foliose lichen *Parmotrema* sp. and is 550 µm long. The tardigrade has attacked the nematode (which appears to be a female enoplid) in the middle of the body. It has a characteristic cylindrical pharynx, the posterior portion of which has been partly digested (A. Bird pers. comm.). *Milnesium tardigradum* is an active predator of both nematodes and other tardigrades.

Macrobiotus sp. (Fig 1C)

One specimen (ATS 507/13) collected and mounted by G. Carruthers, east face of Mt Ainsley, ACT, 10 metres below summit. The tardigrade was found in wet moss and lichen. It is holding the nematode in its mouth. The body walls of the nematode are constricted at either end of the tardigrades mouth. The nematode can not be identified.

DISCUSSION

Four genera of tardigrades are known to prey on nematodes - *Milnesium, Macrobiotus, Isohypsibius* and *Hypsibius*. Six species of *Macrobiotus* have been recorded feeding on nematodes. Preliminary results of a survey of 93 macrobiotid tardigrades suggests that all nematophagous species have proportionally wider buccal tubes with widths at least 17% of buccal tube length. They also all possess buccal lamellae. Using these criteria, a further eight tardigrades may be potentially nematophagous. These are *Macrobiotus boetiae* Durante & Maucci, 1979; *M. gildae* Maucci & Durante Pasa, 1980; *M. grandis* Richters, 1911; *M. maucci* Pilato, 1974; *M. psephus* du Bois-Reymond Mareus, 1944; *M. recens* Cuénot, 1932; *M. suaresensis* Horning *et al.*, 1978 and *M. spectabilis* Thulin, 1928. Further field collecting of these potential nematophagous species is needed to test the hypothesis of Hallas & Yeates (1972) that there is a morphological basis for nematode predation by tardigrades. The width of the buccal tube is not correlated to the length of the tardigrade (Ramazzotti & Maucci 1983) nor does there seem to be any relationship between the size of the nematode being attacked and the size of the attacking tardigrade with nematodes three times larger being attacked (Esser 1990). Large vigorous nematodes may be able to squirm free although observations show that tardigrades can be quite dogged in their attempts to subdue their prey often hanging on for hours (Esser 1990).

It appears that there are many factors involved in nematode prey selection by tardigrades. Hallas & Yeates (1972) have shown a direct correlation between numbers of *Macrobiotus harmsworthi* and the availability of prey, even when environmental factors are taken into account (simple rank correlation + 0.273; moisture excluded = 0.287; temperature excluded + 0.286). When Esser (1990) placed several large populations of tardigrades on water agar containing numerous nematodes of many genera, no feeding or attacking was noted for several weeks. Esser (1990) suggested that the rarity of attacks in agar solution was because the conditions under which tardigrades feed are rarely obtained outside their normal habitat. There also appears to be no correlation between what types of nematodes are eaten by tardigrades. Bacteriophagous, phytophagous and a variety of predacious nematodes have all been taken. One of the problems

in ascertaining what types of nematodes become prey is that the feeding action of tardigrades normally dissolves vital taxonomic features of the nematodes concerned. Tardigrade/nematode interactions occur in a variety of microhabitats and under a range of environmental conditions. Further research needs to be undertaken to understand the roles that these various factors play in both prey selection by tardigrades and the frequency and cause of attacks.

ACKNOWLEDGEMENTS

I wish to thank Glen Carruthers, Melbourne and Sandra Claxton, Macquarie University who made specimens available for this study. Donald Horning, Tumblegum Research Laboratory, Loomberah, New South Wales, Australia and the late Alan Bird, Mitcham, South Australia provided encouragement to publish the work as well as commenting on various drafts of the manuscript.

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Manuscript received 1 August 1999
Revision accepted 8 January 2003